

pMC9 / Tag

tag No. _____

Sample: pMC9 amplification, using the primers # 2721 & 2729

tag dilution: buffers / Klenow

Deprotein

cycling:

10 μM dNTP

1 μM primer each

2 μM Klenow pMC9/Tag II

2 mM Mg

94° 3'

80 (94, 30, 56, 30, 72, 3)

prepared 15 x w/o enzyme → added repeatedly in 1x buffer

x buffer (D.V.)	75	(K7) 75	5	50 μl	1 ml
dNTP	15	15	2.5		.5
buffer 1	7.5	7.5	2		2
4	2	7.5	1.5		1.5
Mg		15.0	1	10 μl	1
Enzyme	3.0	3.0	.5		.5
H ₂ O	642.0	627.0	0		0

distributed 50 μl / tube added enzyme.

0	1	2	3	4	5	6	7	8	9	10	11	12	13	14
		16	17											
			18	19										
				20	21									
					22	23								
						24	25							
							26	27						
								28	29					

0,01 15

30

7

Deprotein mix

1P

rep. Venti buffer Klenow buffer

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Read & Understood by me,

Date

Invent'd by

Date

Recorded by

11/22/84

K. Liberman

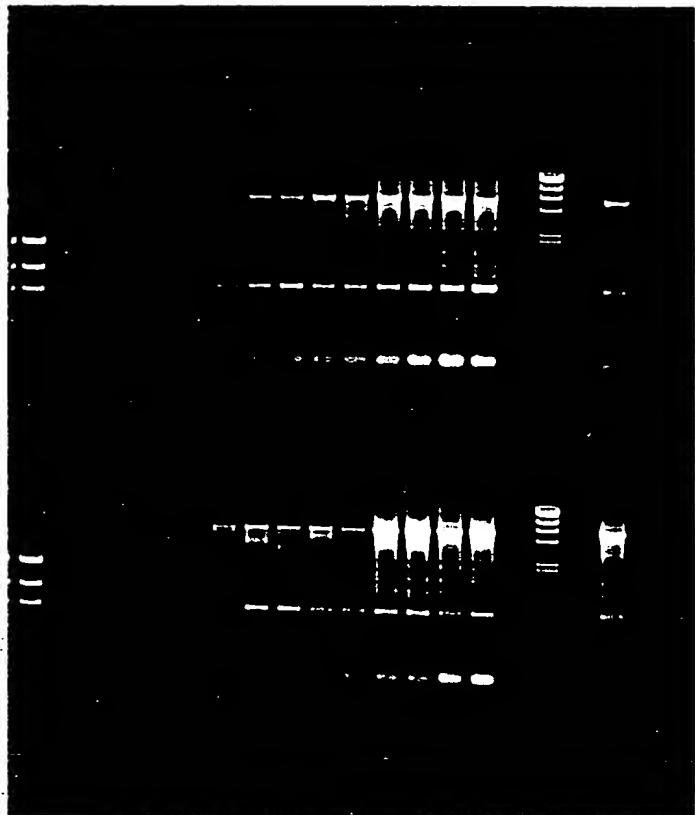
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0 . 5 1 1.5 2 2.5 5

0 . 5 1 1.5 2 5
2.5 1:0.01
max

Tag Kt alone

← D.V buffer

← K.T. buffer

Result: more product with increasing amount of Tag expected.

- K.T. B / I.U. buffer than D.V / I.U.

1: 0.01 but better than I.U. Tag alone.

- K.T. B more product than D.V. buffer.

But lot of misreading - adjust the cycling conditions

T Page No. _____

Witnessed & Understood by me,

Date

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K. Subrahmanian

11/22/94